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DATE: Thursday, September 15, 2005

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☐	L23	L22 and l20	63
☐	L22	l19.ti,ab,clm.	576
☐	L21	L19 with l1 not l17	191
☐	L20	L19 same l1 not l17	619
☐	L19	botulin$ or tetan$	6116
☐	L18	l17 not l11	130
☐	L17	L16 with l1	133
☐	L16	clostrid$ or neurotoxin	7518
☐	L15	L14 same l13 same l9 not l10	5
☐	L14	l4 same (l5 or l6)	17929
☐	L13	l1 same L12	5561
☐	L12	"single chain"	12502
☐	L11	l3 and l10	35
☐	L10	l4 same l5 same l6 same L9	45
☐	L9	cleav$ or protease or proteinase	108341
☐	L6	endocyt$6	4351
☐	L5	transport$	460448
☐	L4	bind$4	366616
☐	L3	l1 with L2	31832
☐	L2	gene or plasmid or protein	191203
☐	L1	fus$4 or chimer$3	261774

END OF SEARCH HISTORY

18sep05 09:09:27 User208600 Session D1699.1

File 155:MEDLINE(R) 1951-2005/Sep 19 (c) format only 2005 Dialog

Set	Items	Description
S1	6955	DC='B3.300.390.400.200.' (CLOSTRIDIUM)
S2	111321	DC='D12.776.828.' (RECOMBINANT PROTEINS)
S3	193	S1 AND S2
S4	7486	DC='D24.185.926.640.' (NEUROTOXINS)
S5	4423	DC='D24.185.926.123.179.' (BOTULINUM TOXINS)
S6	90	S2 AND S5
S7	37269	'MUTAGENESIS, SITE-DIRECTED'
S8	18657	DC='G5.600.' (MUTAGENESIS)
S9	28	S5 AND S7
S10	5	S5 AND S8 NOT S9
S11	13	S5 AND PRECURSOR
S12	136	S4 AND PRECURSOR NOT S11
S13	45	S7 AND S4 NOT S9
S14	136	S12 NOT S13

Ref	Items	Index-term
E1	4	AU=DOLLWET H H
E2	1	AU=DOLLWET-MACK SUSANNE
E3	0	*AU=DOLLY
E4	1	AU=DOLLY C H
E5	7	AU=DOLLY F R
E6	1	AU=DOLLY J
E7	149	AU=DOLLY J O
E8	9	AU=DOLLY J OLIVER
E9	1	AU=DOLLY JOHN P
E10	1	AU=DOLLY M C
E11	3	AU=DOLLY O
E12	3	AU=DOLLY O J
E13	1	AU=DOLLY OLIVER
E14	2	AU=DOLLY R C
E15	1	AU=DOLMA L
E16	1	AU=DOLMA SONAM

S15	162	E6-E8, E12
S16	48	S15 AND S5
S17	0	S7 AND S16
S18	159426	SUBSTITUT?
S19	2	S18 AND S16
S20	0	S16 AND PRECURSOR

6/6/1 18317639 PMID: 15721769
Preparation of specifically activatable endopeptidase derivatives of Clostridium botulinum toxins type A, B, and C and their applications. Mar 2005

6/6/2 18119249 PMID: 15769746
Lipid raft association of SNARE proteins regulates exocytosis in PC12 cells. May 20 2005

6/6/3 17892264 PMID: 15781237
N-terminal helix reorients in recombinant C-fragment of Clostridium botulinum type B. Apr 29 2005

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Simvastatin inhibits growth factor expression and modulates profibrogenic markers in lung fibroblasts. Apr 2005

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Endocytosis and retrograde axonal traffic in motor neurons. 2005

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Use of biophysical characterization in preformulation development of a heavy-chain fragment of botulinum serotype B: evaluation of suitable purification process conditions. Aug 2004

6/6/7 16158501 PMID: 15465919
Using fluorescent sensors to detect botulinum neurotoxin activity in vitro and in living cells. Oct 12 2004

6/6/8 15414141 PMID: 15123599
Synaptotagmins I and II act as nerve cell receptors for botulinum neurotoxin G. Jul 16 2004

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Differential effects of Rho GTPases on axonal and dendritic development in hippocampal neurones. Jul 2004

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RalA-exocyst interaction mediates GTP-dependent exocytosis. May 7 2004

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Plasma membrane localization signals in the light chain of botulinum neurotoxin. Mar 2 2004

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Cloning,high-level expression, single-step purification, and binding activity of His6-tagged recombinant type B botulinum neurotoxin heavy chain transmembrane and binding domain. Mar 2004

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The HCC-domain of botulinum neurotoxins A and B exhibits a singular ganglioside binding site displaying serotype specific carbohydrate interaction. Feb 2004

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Scale-up of the fermentation and purification of the recombinant heavy chain fragment C of botulinum neurotoxin serotype F, expressed in Pichia pastoris. Nov 2003

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Entrapment of Rho ADP-ribosylated by Clostridium botulinum C3 exoenzyme in the Rho-guanine nucleotide dissociation inhibitor-1 complex. Aug 1 2003

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Sequence of the gene for Clostridium botulinum type B neurotoxin associated with infant botulism, expression of the C-terminal half of heavy chain and its binding activity. Jan 3 2003

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Thermal stabilization of the catalytic domain of botulinum neurotoxin E by phosphorylation of a single tyrosine residue. Feb 20 2001

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[Effective expression of fragments of a botulinum neurotoxin type A gene, coding for the L-chain and H-chain in E. coli, with formation of products causing protective immunity to administration of the toxin] Effektivnaia ekspressiia v kletkakah E. coli fragmentov gena botulinicheskogo neirotoksina tipa A, kodiruiushchikh L-tsep' i H-tsep', s obrazovaniem produktov, vyzyvaiushchikh protektivnyi immunitet k vvedeniiu toksina. 2000

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Recognition of RhoA by Clostridium botulinum C3 exoenzyme. Jun 2 2000

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Tyrosine phosphorylation of the vascular endothelial-growth-factor receptor-2 (VEGFR-2) is modulated by Rho proteins. Jun 1 2000

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Fermentation, purification, and efficacy of a recombinant vaccine candidate against botulinum neurotoxin type F from Pichia pastoris. Apr 2000

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Production and purification of the heavy-chain fragment C of botulinum neurotoxin, serotype B, expressed in the methylotrophic yeast Pichia pastoris. Aug 1998

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Regulation of TNF-alpha-induced reorganization of the actin cytoskeleton and cell-cell junctions by Rho, Rac, and Cdc42 in human endothelial cells. Jul 1998

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Botulinum neurotoxin types A and E require the SNARE motif in SNAP-25 for proteolysis. Nov 24 1997

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The interaction of synaptic vesicle-associated membrane protein/synaptobrevin with botulinum neurotoxins D and F. Jun 16 1997

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Analysis of a yeast SNARE complex reveals remarkable similarity to the neuronal SNARE complex and a novel function for the C terminus of the SNAP-25 homolog, Sec9. Jun 27 1997

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Sphingosine 1-phosphate stimulates rho-mediated tyrosine phosphorylation of focal adhesion kinase and paxillin in Swiss 3T3 fibroblasts. Jun 1 1997

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Involvement of Rho and Rac small G proteins and Rho GDI in Ca2+-dependent exocytosis from PC12 cells. Oct 1996

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ADP-ribosylation of the GTP-binding protein RhoA blocks cytoplasmic division in human myelomonocytic cells. Jun 15 1995

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Activation of Clostridium botulinum C3 exoenzyme-catalyzed ADP-ribosylation of RhoA by K+ in a Mg2+ -dependent manner. Jan 1996

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Structural determinants of the specificity for synaptic vesicle-associated membrane protein/synaptobrevin of tetanus and botulinum type B and G neurotoxins. Aug 23 1996

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Directing antigen specificity towards botulinum neurotoxin with combinatorial phage display libraries. Jun 21 1996

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Evidence that syntaxin 1A is involved in storage in the secretory pathway. May 10 1996

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Botulinum neurotoxin C1 cleaves both syntaxin and SNAP-25 in intact and permeabilized chromaffin cells: correlation with its blockade of catecholamine release. Feb 27 1996

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The high-affinity binding of Clostridium botulinum type B neurotoxin to synaptotagmin II associated with gangliosides GT1b/GD1a. Jan 15 1996

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Active site mutation of the C3-like ADP-ribosyltransferase from Clostridium limosum--analysis of glutamic acid 174. Jan 9 1996

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Purification and assay of recombinant C3 transferase. 1995

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Synergistic activation of rat brain phospholipase D by ADP-ribosylation factor and rhoA p21, and its inhibition by Clostridium botulinum C3 exoenzyme. Oct 27 1995

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The t-SNAREs syntaxin 1 and SNAP-25 are present on organelles that participate in synaptic vesicle recycling. Feb 1995

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IgA protease from Neisseria gonorrhoeae inhibits exocytosis in bovine chromaffin cells like tetanus toxin. Jan 27 1995

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Botulinum C3 exoenzyme blocks the tyrosine phosphorylation of p125FAK and paxillin induced by bombesin and endothelin. Nov 14 1994

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The small GTP-binding protein Rho regulates a phosphatidylinositol 4-phosphate 5-kinase in mammalian cells. Nov 4 1994

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A post-docking role for synaptobrevin in synaptic vesicle fusion. Jun 1994

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ADP-ribosylation of Rho proteins by Clostridium botulinum exoenzyme C3 is influenced by phosphorylation of Rho-associated factors. May 15 1994

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Proteolysis of SNAP-25 by types E and A botulinal neurotoxins. Jan 21 1994

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Botulinum neurotoxins serotypes A and E cleave SNAP-25 at distinct COOH-terminal peptide bonds. Nov 29 1993

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Enhancement of Clostridium botulinum C3-catalysed ADP-ribosylation of recombinant rhoA by sodium dodecyl sulfate. Apr 6 1993

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Inhibition of PMA-induced, LFA-1-dependent lymphocyte aggregation by ADP ribosylation of the small molecular weight GTP binding protein, rho. Mar 1993

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ADP-ribosylation by Clostridium botulinum C3 exoenzyme increases steady-state GTPase activities of recombinant rhoA and rhoB proteins. Feb 3 1992

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Identif ication of rho as a substrate for botulinum toxin C3-catalyzed ADP-ribosylation. Apr 24 1989

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The rho gene product expressed in E. coli is a substrate of botulinum ADP-ribosyltransferase C3. Jan 16 1989

6/5/48 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
12482093 PMID: 9792657
Characterization of the catalytic site of the ADP-ribosyltransferase Clostridium botulinum C2 toxin by site-directed mutagenesis.
Barth H; Preiss J C; Hofmann F; Aktories K
Institut fur Pharmakologie und Toxikologie der Albert-Ludwigs-Universitat Freiburg, D-79104 Freiburg, Germany.
Journal of biological chemistry (UNITED STATES) Nov 6 1998, 273 (45) p29506-11, ISSN 0021-9258 Journal Code: 2985121R Publishing Model Print
Document type: Journal Article Languages: ENGLISH Main Citation Owner: NLM
Record type: MEDLINE; Completed Subfile: INDEX MEDICUS
The actin ADP-ribosylating Clostridium botulinum C2 toxin is a binary toxin composed of the binding component C2II and the enzyme component C2I. C2I ADP-ribosylates G-actin at arginine 177, resulting in the depolymerization of the actin cytoskeleton. Here, we studied the structure-function relationship of C2I by site-directed mutagenesis. Exchange of Glu389 to glutamine caused the complete loss of ADP-ribosyltransferase and NAD-glycohydrolase activities of C2I. In contrast, exchange of Glu387 to glutamine blocked ADP-ribosyltransferase but not NAD-glycohydrolase activity. Whereas photoaffinity labeling of the double mutant E387Q/E389Q C2I with [carbonyl-14C]NAD was blocked, labeling of the single C2I mutants was reduced (E389Q) or not changed (E387Q). Exchange of the STS motif (amino acid residues 348-350) of C2I caused a decrease in transferase activity by more than 99 (S348A) and 90% (T349V), or did not affect activity (S350A). Exchange of Arg299 and Arg300 to lysine reduced transferase activity to <0.1 and

approximately 35% of wild-type activity. The data indicate that the amino acid residues Glu389, Glu387, Ser348, and Arg299, which are conserved in various prokaryotic and eukaryotic arginine-modifying ADP-ribosyltransferases, are essential for ADP-ribosyltransferase activity of the enzyme component of C. botulinum C2 toxin.

Tags: Research Support, Non-U.S. Gov't

Descriptors: *Botulinum Toxins--metabolism--ME; *Poly(ADP-ribose) Polymerases--metabolism--ME; Amino Acid Sequence; Animals; Botulinum Toxins--chemistry--CH; Botulinum Toxins--genetics--GE; CHO Cells; Catalytic Domain; Hamsters; Humans; Molecular Sequence Data; Mutagenesis, Site-Directed; Photoaffinity Labels; Poly(ADP-ribose) Polymerases--chemistry--CH; Poly(ADP-ribose) Polymerases--genetics--GE; Recombinant Proteins--chemistry--CH; Recombinant Proteins--genetics--GE; Recombinant Proteins--metabolism--ME; Sequence Homology, Amino Acid CAS Registry No.: 0 (Botulinum Toxins); 0 (Photoaffinity Labels); 0 (Recombinant Proteins); 0 (botulinum toxin type C)

Enzyme No.: EC 2.4.2.30 (Poly(ADP-ribose) Polymerases)

Record Date Created: 19981210 Record Date Completed: 19981210

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12383413 PMID: 9693060

Production and purification of the heavy-chain fragment C of botulinum neurotoxin, serotype B, expressed in the methylotrophic yeast Pichia pastoris.

Potter K J; Bevins M A; Vassilieva E V; Chiruvolu V R; Smith T; Smith L A ; Meagher M M

Department of Food Science and Technology, Biological Process Development Facility, University of Nebraska-Lincoln, 68583-0919, USA.

Protein expression and purification (UNITED STATES) Aug 1998, 13 (3) p357-65, ISSN 1046-5928 Journal Code: 9101496 Publishing Model Print Document type: Journal Article Languages: ENGLISH Main Citation Owner: NLM

Record type: MEDLINE; Completed Subfile: INDEX MEDICUS

A recombinant Hc fragment of botulinum neurotoxin, serotype B (rBoNTB(Hc)), has been successfully expressed in a Mut+ strain of the methylotrophic yeast Pichia pastoris for use as an antigen in a proposed human vaccine. The fermentation process consisted of batch phase on glycerol, followed by glycerol and methanol fed-batch phases yielding a final cell mass of 60 g/L (dcw) and was easily scaled-up to 60 L. A multistep ion-exchange chromatographic purification process was employed to produce 99% pure Hc fragment. The final yield of the purified antigen was 390 mg per kilogram of wet cell mass. The purified Hc fragment of serotype B was stable, elicited an immune response in mice, and protected upon challenge with native botulin. Copyright 1998 Academic Press.

Tags: Research Support, U.S. Gov't, Non-P.H.S.

Descriptors: *Botulinum Toxins--genetics--GE; *Neurotoxins--genetics--GE; *Pichia--genetics--GE; Amino Acid Sequence; Animals; Blotting, Western; Botulinum Toxins--chemistry--CH; Botulinum Toxins--isolation and purification--IP; Chromatography, Ion Exchange; Cloning, Molecular; Electrophoresis, Polyacrylamide Gel; Mice; Molecular Sequence Data; Neurotoxins--chemistry--CH; Neurotoxins--isolation and purification--IP; Recombinant Proteins--chemistry--CH; Recombinant Proteins--genetics--GE; Recombinant Proteins--isolation and purification--IP CAS Registry No.: 0 (Botulinum Toxins); 0 (Neurotoxins); 0 (Recombinant Proteins)

Record Date Created: 19980917 Record Date Completed: 19980917

9/6/1 18520371 PMID: 15938619

Analysis of active site residues of botulinum neurotoxin E by mutational, functional, and structural studies: Glu335Gln is an apoenzyme. Jun 14 2005

9/6/2 18119249 PMID: 15769746

Lipid raft association of SNARE proteins regulates exocytosis in PC12 cells. May 20 2005

9/6/3 15324488 PMID: 14978027

RalA -exocyst interaction mediates GTP-dependent exocytosis. May 7 2004

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Structural analysis by X-ray crystallography and calorimetry of a haemagglutinin component (HA1) of the progenitor toxin from Clostridium botulinum. Dec 2003

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Rho-specific Bacillus cereus ADP-ribosyltransferase C3cer cloning and characterization. Aug 19 2003

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Nitric oxide-induced decrease in calcium sensitivity of resistance arteries is attributable to activation of the myosin light chain phosphatase and antagonized by the RhoA/Rho kinase pathway. Jun 24 2003

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Channel formation by the binding component of Clostridium botulinum C2 toxin: glutamate 307 of C2II affects channel properties in vitro and pH-dependent C2I translocation in vivo. May 13 2003

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The core membrane fusion complex governs the probability of synaptic vesicle fusion but not transmitter release kinetics. Feb 15 2002

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Structure-function analysis of the Rho-ADP-ribosylating exoenzyme C3stau2 from Staphylococcus aureus. Feb 5 2002

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SNAP -25 with mutations in the zero layer supports normal membrane fusionkinetics. Dec 2001

9/6/11 13946422 PMID: 11700044

Site-directed mutagenesis identifies active-site residues of the light chain of botulinum neurotoxin type A. Nov 16 2001

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A discontinuous SNAP-25 C-terminal coil supports exocytosis. Jul 27 2001

9/6/13 13687573 PMID: 11329292

Thermal stabilization of the catalytic domain of botulinum neurotoxin E by phosphorylation of a single tyrosine residue. Feb 20 2001

9/6/14 13520056 PMID: 10488092

Galpha(13) stimulates Rho-dependent activation of the cyclooxygenase-2 promoter. Sep 24 1999

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Protei n kinase C inhibits Kv1.1 potassium channel function. Jul 1999

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SNAP -25a and -25b isoforms are both expressed in insulin-secreting cells and can function in insulin secretion. Apr 1 1999

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Memb rane localization and biological activity of SNAP-25 cysteine mutants in insulin-secreting cells. Sep 2000

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Recognition of RhoA by Clostridium botulinum C3 exoenzyme. Jun 2 2000

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Rho-A is critical for osteoclast podosome organization, motility, and bone resorption. Apr 21 2000

9/6/20 12660268 PMID: 10582602

Botulinum neurotoxin E-insensitive mutants of SNAP-25 fail to bind VAMP but support exocytosis. Dec 1999

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Chara cterization of the catalytic site of the ADP-ribosyltransferase Clostridium botulinum C2 toxin by site-directed mutagenesis. Nov 6 1998

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The small GTP-binding protein RacG regulates uroid formation in the protozoan parasite Entamoeba histolytica. Jun 1998

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Botulinum neurotoxin types A and E require the SNARE motif in SNAP-25 for proteolysis. Nov 24 1997

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The interaction of synaptic vesicle-associated membrane protein/synaptobrevin with botulinum neurotoxins D and F. Jun 16 1997

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Rho proteins play a critical role in cell migration during the early phase of mucosal restitution. Jul 1 1997

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Mutational analysis of VAMP domains implicated in Ca2+-induced insulin exocytosis. Dec 16 1996

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Active site mutation of the C3-like ADP-ribosyltransferase from Clostridium limosum--analysis of glutamic acid 174. Jan 9 1996

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Identification of Glu173 as the critical amino acid residue for the ADP-ribosyltransferase activity of Clostridium botulinum C3 exoenzyme. Sep 4 1995

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The C terminus of component C2II of Clostridium botulinum C2 toxin is essential for receptor binding. Aug 2000

10/6/2 12469820 PMID: 9782457

On the action of botulinum neurotoxins A and E at cholinergic terminals. Apr 1998

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Ganglioside GT1b as a complementary receptor component for Clostridium botulinum neurotoxins. Aug 1998

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Expre ssion and purification of the light chain of botulinum neurotoxin A e single mutation abolishes its cleavage of SNAP-25 and neurotoxicity after reconstitution with the heavy chain. Nov 21 1995

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ADP-ribosylation of Drosophila indirect-flight-muscle actin and arthrin by Clostridium botulinum C2 toxin and Clostridium perfringens iota toxin. Apr 15 1993

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Lysop hosphatidic acid induces process retraction in CG-4 line oligodendrocytes and oligodendrocyte precursor cells but not in differentiated oligodendrocytes. Nov 2003

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[Botulism: the agent, mode of action of the botulinum neurotoxins, forms of acquisition, treatment and prevention] Le botulisme: agent, mode d'action des neurotoxines botuliques, formes d'acquisition, traitement et prevention. Aug 2002

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Botulinum neurotoxin types B and E: purification, limited proteolysis by endoproteinase Glu-C and pepsin, and comparison of their identified cleaved sites relative to the three-dimensional structure of type A neurotoxin.0 Oct 2001

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Tetanus and botulism neurotoxins: a new group of zinc proteases. Sep 1993

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Circular dichroic and fluorescence spectroscopic study of the conformation of botulinum neurotoxin types A and E. Feb 1988

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Molecular characterization of a protein, insoluble at low temperature, produced by Clostridium botulinum type G. Aug 1986

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Purification and amino acid composition of type A botulinum neurotoxin. 1984

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[Characteristics of the activation of type E. Cl. botulinum toxins] Nekotorye osobennosti aktivatsii botulinicheskogo toksina tipa E. Apr 1982

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[Isolation and properties of highly purified C1. botulinum toxin type E] Vydelenie i nekotorye svoistva vysokoochishchennogo botulinicheskogo toksina tipa E. Aug 1979

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13812149 PMID: 11478959
Botulinum neurotoxin types B and E: purification, limited proteolysis by endoproteinase Glu-C and pepsin, and comparison of their identified cleaved sites relative to the three-dimensional structure of type A neurotoxin.
Prabakaran S; Tepp W; DasGupta B R
Department of Food Microbiology and Toxicology, University of Wisconsin, Madison, 53706, USA.
Toxicon - official journal of the International Society on Toxinology (England) Oct 2001, 39 (10) p1515-31, ISSN 0041-0101 Journal Code: 1307333
Contract/Grant No.: NS 17742; NS; NINDS Publishing Model Print
Document type: Journal Article Languages: ENGLISH Main Citation Owner: NLM
Record type: MEDLINE; Completed
Botulinum neurotoxin (NT) serotypes B and E are approximately 150 kDa proteins. Isolated from the liquid culture of Clostridium botulinum the NT type E is a single chain protein while the NT type B, from the proteolytic strain of the bacteria, is a mixture of dichain (nicked within a disulfide loop located about one-third the way from the N-terminus to the C-terminus) protein and its precursor single-chain protein. Endoproteinase Glu-C (EC 3.4.21.19) and pepsin (EC 3.4.23.1) were used for controlled digestion of NT types B and E; the amino acid residues flanking many of the cleavable peptide bonds were identified and the corresponding proteolytic fragments partially characterized. Chemical identification of 82 and 108 residues of types B and E NT, respectively, revealed that the residue 738 and 1098 in type E NT, identified as Leu and Asn, respectively, differ from those deduced from nucleotide sequences. Several fragments overlapped spanning various segments of the NT's functional domains; they appear to have potential for structure-function studies of the NT. The cleavage sites were compared with the previously determined proteolyzed sites on NT types A and E. The cleavage sites of the NT types A, B and E, all exposed on the protein surface, were scrutinized in the context of the three-dimensional structure of crystallized NT type A [Lacy, D.B., Stevens, R.C., 1999. J. Mol. Biol. 291, 1091-1104]. Detailed procedures for isolation of pure NT types B and E in large quantities (average yield 92 and 62 mg, respectively) suitable for crystallization are reported.
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11/5/8 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
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Tetanus and botulism neurotoxins: a new group of zinc proteases.
Montecucco C; Schiavo G
Department of Biomedical Sciences, University of Padova, Italy.
Trends in biochemical sciences (ENGLAND) Sep 1993, 18 (9) p324-7, ISSN 0968-0004 Journal Code: 7610674
Publishing Model Print Document type: Journal Article; Review; Review, Tutorial
Languages: ENGLISH
Main Citation Owner: NLM Record type: MEDLINE; Completed Subfile: INDEX MEDICUS
The active forms of tetanus and botulinum neurotoxins, released from the precursor molecule by specific proteolysis and reduction, block the release of neurotransmitters via a Zn(2+)-dependent protease activity. VAMP/synaptobrevin, an integral membrane protein of the synaptic vesicles, is cleaved at a single site by tetanus and botulinum B, D and F neurotoxins. The unique sequence, mechanism of activation and site of activity of clostridial neurotoxins mark them out as an independent group of Zn(2+)-endopeptidases. (38 Refs.)
Tags: Research Support, Non-U.S. Gov't
CAS Registry No.: 0 (Botulinum Toxins); 0 (Membrane Proteins); 0 (Nerve Tissue Proteins); 0 (Neurotransmitters); 0 (Tetanus Toxin); 0 (vesicle-associated membrane protein)
Enzyme No.: EC 3.4.24 (Metalloendopeptidases); EC 3.4.24.- (zinc-endopeptidase, tetanus neurotoxin)
Record Date Created: 19931214 Record Date Completed: 19931214

11/7/10 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
07740340 PMID: 3538716
Molecular characterization of a protein, insoluble at low temperature, produced by Clostridium botulinum type G.
Gimenez J A; Cascone O; Biscoglio de Jimenez Bonino M J; Paladini A C
Zentralblatt fur Bakteriologie, Mikrobiologie, und Hygiene. Series A, Medical microbiology, infectious diseases, virology, parasitology (GERMANY, WEST) Aug 1986, 262 (2) p179-88, ISSN 0176-6724 Journal Code: 8403032
Publishing Model Print Document type: Journal Article Languages: ENGLISH
Main Citation Owner: NLM
Record type: MEDLINE; Completed
A preliminary study of a low-toxicity protein, called cryoprotein, produced by Clostridium botulinum type G, led to a better characterization of this substance and to discriminate its relationship with type G botulinum toxin. This sparingly soluble protein has been characterized as an aggregated form of a soluble precursor with an Mr of 170,000. This phenomenon is temperature-dependent. The monomeric protein is usually contaminated with a lower Mr form (150,000) quite probably originated by a limited proteolytic process. The amino acid composition of this protein is relatively analogous to that of the botulinum toxins A and B, the only notable exception being the absence of cysteine. The N-terminal amino acid is alanine and the C-terminal sequence is Val-Ala-Leu-OH. The low toxicity which is usually demonstrable in samples of this protein disappears after a reductive treatment, strongly suggesting that it is not an intrinsic property. Taking into account that some of its physiochemical properties are similar to those of the known botulinal toxins, it is quite probable that this substance accompanies G toxin preparations currently obtained by routine methods, increasing its non-toxic antigenic mass. This fact could be critical to the sensitivity and specificity of G toxin immunological detection methods.
Record Date Created: 19870102 Record Date Completed: 19870102

11/7/11 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
07042616 PMID: 6382680
Purification and amino acid composition of type A botulinum neurotoxin.
DasGupta B R; Sathyamoorthy V
Toxicon - official journal of the International Society on Toxinology (ENGLAND) 1984, 22 (3) p415-24, ISSN 0041-0101 Journal Code: 1307333 Contract/Grant No.: NS17742; NS; NINDS Publishing Model Print Document type: Journal Article
Languages: ENGLISH Main Citation Owner: NLM Record type: MEDLINE; Completed
A method to purify type A botulinum neurotoxin from a 64 liter bacterial culture is reported. The procedure includes cation exchange chromatography at pH 7.0. The final product, essentially homogeneous (according to polyacrylamide gel-sodium dodecylsulfate electrophoresis), is a mixture of two forms of the neurotoxin (mol. wt 145,000); the dichain or nicked form (over 95%) and its precursor the single chain or unnicked form. Two batches of the neurotoxin purified by the method described here and one batch purified according to the method of Sugii and Sakaguchi were similar in purity and amino acid composition. The best estimate of the number of amino acid residues per neurotoxin molecule (mol. wt 145,000) is:
Asp200Thr75Ser79Glu114Pro44Gly64Ala53Val70CyS10Met22Ile111Leu104Tyr71 Phe68 Lys100His14Arg43Trp17.
Record Date Created: 19841012 Record Date Completed: 19841012

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A spider toxin that induces a typical effect of scorpion alpha-toxins but competes with beta-toxins on binding to insect sodium channels. Feb 8 2005

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Topology of the substrate-binding site of a Lys49-phospholipase A2 influences Ca2+-independent membrane-damaging activity. Aug 15 2004

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Basic amino acid residues in the beta-structure region contribute, but not critically, to presynaptic neurotoxicity of ammodytoxin A. Nov 1 2004

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Conversion of a scorpion toxin agonist into an antagonist highlights an acidic residue involved in voltage sensor trapping during activation of neuronal Na+ channels. Apr 2004

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Resiniferatoxin binds to the capsaicin receptor (TRPV1) near the extracellular side of the S4 transmembrane domain. Mar 9 2004

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The effects of in vitro application of purified botulinum neurotoxin at mouse motor nerve terminals. May 1987

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Localization of sites for 125I-labelled botulinum neurotoxin at murine neuromuscular junction and its binding to rat brain synaptosomes. 1982

19/7/1 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
11179774 PMID: 7578132
Expression and purification of the light chain of botulinum neurotoxin A: a single mutation abolishes its cleavage of SNAP-25 and neurotoxicity after reconstitution with the heavy chain.
Zhou L; de Paiva A; Liu D; Aoki R; Dolly J O
Department of Biochemistry, Imperial College of Science, Technology and Medicine, London.
Biochemistry (UNITED STATES) Nov 21 1995, 34 (46) p15175-81, ISSN 0006-2960

Journal Code: 0370623
Publishing Model Print Document type: Journal Article Languages: ENGLISH Main Citation Owner: NLM
Record type: MEDLINE; Completed
Botulinum neurotoxin type A (BoNT/A) selectively and irreversibly inhibits acetylcholine release from peripheral nerve endings. While the toxin's heavy (H) chain contributes to neuronal binding and internalization, its light (L) chain is a Zn(2+)-dependent endoprotease that intracellularly cleaves synaptosomal-associated protein of M(r) = 25 kDa (SNAP-25). For research and clinical exploitation of this uniquely-acting neurotoxin, recombinant wild-type L chain was produced together with a mutant in which His227 in the Zn(2+)-binding motif was substituted by Tyr. The PCR-amplified wild-type and mutant L chain genes were cloned, fused to the gene for maltose-binding protein, and expressed at high levels in Escherichia coli. The soluble fusion proteins were purified using amylose affinity chromatography, and, after factor Xa cleavage, the free L chains were isolated. The wild-type was shown to proteolyze SNAP-25 at a rate approaching that of the native chain while the mutant was inactive. Reconstitution of the pure wild-type L chain with native homogeneous H chain yielded a disulfide-linked dichain form that inhibited neuromuscular transmission in vitro and produced the symptoms of botulism in vivo. After reconstitution with the H chain, the Tyr227 mutant L chain failed to show any neuroparalytic activity in either of these assays. This methodology allows, for the first time, routine preparation of recombinant forms of the L chain that are needed to decipher the molecular details of its interaction with substrate and, thereby, assist the design of effective inhibitors.(ABSTRACT TRUNCATED AT 250 WORDS)
Record Date Created: 19951226 Record Date Completed: 19951226

19/7/2 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
09931419 PMID: 1356988
Tetanus toxin and botulinum toxins type A and B inhibit glutamate, gamma-aminobutyric acid, aspartate, and met-enkephalin release from synaptosomes. Clues to the locus of action.
McMahon H T; Foran P; Dolly J O ; Verhage M; Wiegant V M; Nicholls D G
Department of Biochemistry, University of Dundee, United Kingdom.
Journal of biological chemistry (UNITED STATES) Oct 25 1992, 267 (30) p21338-43, ISSN 0021-9258 Journal Code: 2985121R Publishing Model Print Document type: Journal Article Languages: ENGLISH
Main Citation Owner: NLM Record type: MEDLINE; Completed
Tetanus toxin (100 nM) when preincubated with guinea pig cerebrocortical synaptosomes for 45 min reduces the final extent of the KCl-evoked, Ca(2+)-dependent, glutamate transmitter release to 30% of non-intoxicated controls. Similarly, 100 nM Botulinum neurotoxins, types A and B, preincubated for 90 min inhibit release to 45-60% of non-intoxicated controls. The toxins preferentially attenuate a slow phase of KCl-evoked glutamate release which may be associated with synaptic vesicle mobilization. Tetanus toxin additionally inhibits the release of aspartate, gamma-aminobutyric acid and met-enkephalin from the same preparation. Since amino acids and neuropeptides are released by distinct mechanisms, this indicates that the toxin affects a step common to both exocytotic pathways. When Ba2+ (which does not interact with calmodulin) is substituted for Ca2+, the control KCl-evoked release of each transmitter is unaffected and tetanus toxin is still inhibitory. Taken together these results implicate a calmodulin-independent locus (or loci) of action common to small- and large-dense-core vesicles and associated with vesicle transport.
Record Date Created: 19921125 Record Date Completed: 19921125

18sep05 11:05:15 User208600 Session D1700.1

File 155:MEDLINE(R) 1951-2005/Sep 19 (c) format only 2005 Dialog

Set	Items	Description
S1	6955	DC='B3.300.390.400.200.' (CLOSTRIDIUM)
S2	111321	DC='D12.776.828.' (RECOMBINANT PROTEINS)
S3	193	S1 AND S2
S4	7486	DC='D24.185.926.640.' (NEUROTOXINS)
S5	4423	DC='D24.185.926.123.179.' (BOTULINUM TOXINS)
S6	90	S2 AND S5
S7	37269	'MUTAGENESIS, SITE-DIRECTED'
S8	18657	DC='G5.600.' (MUTAGENESIS)
S9	28	S5 AND S7
S10	5	S5 AND S8 NOT S9
S11	13	S5 AND PRECURSOR
S12	1771	DC='D24.185.926.123.893.' (TETANUS TOXIN)
S13	11	S7 AND S12
S14	13	S12 AND PRECURSOR
S15	4	S8 AND S12 NOT S13

13/6/1 14641734 PMID: 12556363
Tetanus toxin abolishes exocytosis of ROMK1 induced by inhibition of protein tyrosine kinase. Mar 2003

13/6/2 14483514 PMID: 12427026
Identification of a binding site for ganglioside on the receptor binding domain of tetanus toxin. Nov 19 2002

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Site-directed mutagenesis identifies active-site residues of the light chain of botulinum neurotoxin type A. Nov 16 2001

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Recombinant forms of tetanus toxin engineered for examining and exploiting neuronal trafficking pathways. Aug 17 2001

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Tyrosine-1290 of tetanus neurotoxin plays a key role in its binding to gangliosides and functional binding to neurones. Mar 23 2001

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Control of antigen presentation by a single protease cleavage site. Apr 2000

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Adjuvant effect of non-toxic mutants of E. coli heat-labile enterotoxin following intranasal, oral and intravaginal immunization. 1998

13/6/9 10722987 PMID: 7929330
Cytotoxic effects of a chimeric protein consisting of tetanus toxin light chain and anthrax toxin lethal factor in non-neuronal cells. Oct 21 1994

13/6/10 10607472 PMID: 7911329
A single mutation in the recombinant light chain of tetanus toxin abolishes its proteolytic activity and removes the toxicity seen after reconstitution with native heavy chain. Jun 7 1994

13/6/11 10186582 PMID: 8500613
Production of biologically active light chain of tetanus toxin in Escherichia coli. Evidence for the importance of the C-terminal 16 amino acids for full biological activity. Jun 1 1993

13/7/7 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
12853467 PMID: 10795737
Control of antigen presentation by a single protease cleavage site.
Antoniou A N; Blackwood S L; Mazzeo D; Watts C
Department of Biochemistry, Wellcome Trust Biocentre, University of Dundee, United Kingdom.
Immunity (UNITED STATES) Apr 2000, 12 (4) p391-8, ISSN 1074-7613 Journal Code: 9432918
Publishing Model Print Document type: Journal Article Languages: ENGLISH
Main Citation Owner: NLM Record type: MEDLINE; Completed
Protein antigens require limited proteolytic processing to generate peptides for binding to class II MHC molecules, but the proteases and processing sites involved are largely unknown. Here we analyze the effect of eliminating the three major asparagine endopeptidase (AEP)-processing sites in the microbial antigen tetanus toxin C fragment. The mutant antigen is highly resistant to proteolysis by AEP and crude lysosomal extracts and is dramatically impaired in its ability to be processed and presented to T cells. Remarkably, processing at a single asparagine residue (1219) is obligatory for optimal presentation of many T cell epitopes in this antigen. These studies demonstrate that cleavage at a single processing site can be crucial for effective antigen presentation.
Record Date Created: 20000523 Record Date Completed: 20000523

13/7/9 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
10722987 PMID: 7929330
Cytotoxic effects of a chimeric protein consisting of tetanus toxin light chain and anthrax toxin lethal factor in non-neuronal cells.
Arora N; Williamson L C; Leppla S H; Halpern J L
Laboratory of Microbial Ecology, NIDR, National Institutes of Health, Bethesda, Maryland 20892.
Journal of biological chemistry (UNITED STATES) Oct 21 1994, 269 (42) p26165-71, ISSN 0021-9258 Journal Code: 2985121R Publishing Model Print Document type: Journal Article Languages: ENGLISH
Main Citation Owner: NLM Record type: MEDLINE; Completed
The light chain of tetanus toxin is a zinc endoprotease that inhibits neurotransmitter release by selective proteolysis of the synaptic vesicle-associated protein synaptobrevin/vesicle-associated membrane protein. Cellubrevin is a homologue of synaptobrevin that is found in most cell types and is also a substrate for tetanus toxin. The lack of receptors for tetanus toxin on most cell types has made studies of tetanus toxin action in non-neuronal cells difficult. To characterize tetanus toxin effects in non-neuronal cells, a fusion protein consisting of the 254 amino-terminal amino acids of lethal factor (LF) of anthrax toxin and tetanus toxin light chain (LC) was prepared. This protein (LF-LC) inhibited evoked glycine release from primary spinal cord neurons at concentrations between 1.0 and 100 ng/ml. LF-LC was cytotoxic to RAW 264.7, ANA-1 cells (mouse macrophage cell lines), and Chinese hamster ovary cells in a dose-dependent manner. These effects required the presence of protective antigen, the receptor binding component of anthrax toxin. In contrast, LF-LC was not cytotoxic to RBL-2H3, Vero, or mouse hybridoma cell lines. Mutagenesis of conserved amino acids (His237 and Glu234) in the zinc-binding motif of LC resulted in fusion proteins having no biological activity. LF-LC did not inhibit regulated secretion of serotonin in RBL-2H3 cells or constitutive secretion in any non-neuronal cell lines as measured in several different assays. We suggest that the cytotoxic effects of LF-LC result from inhibition of a specific intracellular membrane fusion event mediated by cellubrevin.
Record Date Created: 19941122 Record Date Completed: 19941122

13/7/10 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
10607472 PMID: 7911329
A single mutation in the recombinant light chain of tetanus toxin abolishes its proteolytic activity and removes the toxicity seen after reconstitution with native heavy chain.
Li Y; Foran P; Fairweather N F; de Paiva A; Weller U; Dougan G; Dolly J O
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Biochemistry (UNITED STATES) Jun 7 1994, 33 (22) p7014-20, ISSN 0006-2960 Journal Code: 0370623
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Specific proteolysis by the tetanus toxin light chain of a vesicle-associated membrane protein (VAMP) involved in exocytosis is thought to underlie its intracellular blockade of neurotransmitter release. To substantiate this mechanism, recombinant light chain was expressed as a maltose binding protein-light chain fusion product in Escherichia coli. After purification of affinity chromatography and cleavage with factor Xa, the resultant light chain was isolated and its identity confirmed by Western blotting and N-terminal sequencing. It exhibited activity similar to that of the native light chain in proteolyzing its target in isolated bovine small synaptic vesicles and in hydrolyzing a 62-residue synthetic polypeptide spanning the cleavage site of the substrate. The importance of Glu234 in the catalytic activity of the light chain, possibly analogous to Glu143 of thermolysin, was examined using site-directed mutagenesis. Changing Glu234 to Ala abolished the protease activity of the light chain, but its ability to bind the polypeptide substrate was retained. Each recombinant light chain could be reconstituted with the heavy chain of tetanus toxin, yielding the same level of disulfide-linked species as the two native chains. Whereas the toxin formed with wild-type light chain exhibited appreciable neuromuscular paralysis activity and mouse lethality, the equivalent dichain material containing the Ala234 mutant lacked neurotoxicity in both the in vitro and in vivo assays. Thus, these results demonstrate directly, for the first time, that the lethality of tetanus toxin and its inhibition of exocytosis in intact neurons are attributable largely, if not exclusively, to endoprotease activity.
Record Date Created: 19940708 Record Date Completed: 19940708

13/7/11 DIALOG(R)File 155:MEDLINE(R) (c) format only 2005 Dialog. All rts. reserv.
10186582 PMID: 8500613
Production of biologically active light chain of tetanus toxin in Escherichia coli. Evidence for the importance of the C-terminal 16 amino acids for full biological activity.

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FEBS letters (NETHERLANDS) Jun 1 1993, 323 (3) p218-22, ISSN 0014-5793 Journal Code: 0155157
Publishing Model Print Document type: Journal Article Languages: ENGLISH Main Citation Owner: NLM
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The activity of the light (L) chain of tetanus toxin, and of mutants constructed by site-directed mutagenesis, was studied by expression and purification of the proteins from E. coli. Wild-type recombinant L chain (pTet87) was active in the inhibition of exocytosis from cultured bovine adrenal chromaffin cells, although at a level 5-15% of that of L chain purified from tetanus toxin. L chain mutants which terminated at Leu-438 (pTet89), or which contained a Cys-to-Ser mutation at residue 439 (pTet88) were equally as active as the full-length recombinant protein. The reduced activity of pTet87 L chain correlated with C-terminal proteolysis of the protein upon purification. A tryptic fragment derived from native light chain and which terminated at Leu-434 also showed reduced activity in the exocytosis assay, consistent with a requirement of the C-terminal region of the L chain for maximal activity. pTet87 L chain, but neither of the mutants, could be associated with purified H (heavy) chain to form a covalent dimer which induced the symptoms of tetanus in mice. The ability to form biologically active toxin using recombinant L chain will be of great value in structure-function studies of tetanus toxin.
Record Date Created: 19930629 Record Date Completed: 19930629

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Characterization of tetanus toxin, neat and in culture supernatant, by electrospray mass spectrometry. Feb 15 2002

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Chains and fragments of tetanus toxin, and their contribution to toxicity. 1990

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Neural induction and in vitro initial expression of neurofilament and tetanus toxin binding site molecules in amphibians. Jan 1986

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In vitro differentiation of neuronal precursor cells from amphibian late gastrulae; morphological, immunocytochemical studies, biosynthesis, accumulation and uptake of neurotransmitters. Apr 1985

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Neuronal acquisition of tetanus toxin binding sites: relationship with the last mitotic cycle. Dec 1983

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Tracing the development of oligodendrocytes from precursor cells using monoclonal antibodies, fluorescence-activated cell sorting, and cell culture. Nov 1983

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Tetanus toxin: a marker of amphibian neuronal differentiation in vitro. Mar 10 1981

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08402736 PMID: 3054567 Record Identifier: 89040247
Tetanus toxin: biochemical and pharmacological comparison between its protoxin and some isotoxins obtained by limited proteolysis.
Weller U; Mauler F; Habermann E
Rudolf-Buchheim-Institut fur Pharmakologie, Justus-Liebig-Universitat Giessen, Federal Republic of Germany.
Naunyn-Schmiedeberg's archives of pharmacology (GERMANY, WEST) Aug 1988 , 338 (2) p99-106, ISSN 0028-1298
Journal Code: 0326264 Publishing Model Print Document type: Journal Article Languages: ENGLISH
Main Citation Owner: NLM Other Citation Owner: NASA Record type: MEDLINE; Completed Subfile: INDEX MEDICUS
Single-chain tetanus toxin (toxin S) was prepared from short-term cultures by lysis under protection with protease inhibitors, precipitation with 40% ammonium sulfate, gel filtration, and chromatography on DEAE ion exchanger. Its limited proteolysis by trypsin, post-arginine cleaving enzyme from mouse submaxillary gland and clostripain led to bichainal derivatives (BT, BA, BCl) consisting of a heavy chain and a larger version of the light chain. The latter was then converted by trypsin into a small version which comigrated with the light chain of bichainal extracellular toxin (BE). The light chain produced by chymotrypsin (BC) and elastase (BE1) was of intermediate size. The nick region serves as substrate for all esteroproteases investigated and comprises between one and two kDa. Limited proteolysis increased the hydrophilicity (BT greater than BE greater than S) in hydrophobic interaction HPLC, and anionic behaviour (BC greater than BE greater than BT greater than S) in DEAE ion exchanger HPLC. The bichainal toxins assessed (BC, BE or BT) were about two times more toxic than toxin S (LD50, mouse s.c. 2 ng/kg vs. 4 ng/kg). They were five to twelve times more potent than toxin S in three in vitro assays measuring the prevention of neurotransmitter release, i.e. on the phrenic nerve-hemidiaphragm preparation of the mouse (acetylcholine, with toxin BE and BT), on primary brain cell cultures from the mouse ([3H]noradrenaline, with toxin BE and BT), and on brain homogenate from rats ([3H]noradrenaline, with toxin BA, BC, BE and BT). Thus single-chain toxin is a less potent precursor of, or protoxin for, various bichainal isotoxins.(ABSTRACT TRUNCATED AT 250 WORDS)
Tags: Comparative Study; In Vitro; Research Support, Non-U.S. Gov't
Descriptors: *Tetanus Toxin--isolation and purification--IP; Animals; Brain--drug effects--DE; Brain--metabolism--ME; Chromatography--methods --MT; Hydrolysis; Mice; Neuromuscular Junction--drug effects--DE; Norepinephrine--metabolism--ME; Peptide Fragments --isolation and purification--IP; Peptide Fragments--metabolism--ME; Peptide Fragments --toxicity--TO; Peptide Hydrolases; Rats; Tetanus Toxin--metabolism--ME; Tetanus Toxin--toxicity--TO
CAS Registry No.: 0 (Peptide Fragments); 0 (Tetanus Toxin); 51-41-2 (Norepinephrine)
Enzyme No.: EC 3.4.- (Peptide Hydrolases)
Record Date Created: 19881221 Record Date Completed: 19881221

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Activation of store-operated calcium channels: assessment of the role of snare-mediated vesicular transport. Aug 15 2003

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The LTR72 mutant of heat-labile enterotoxin of Escherichia coli enhances the ability of peptide antigens to elicit CD4(+) T cells and secrete gamma interferon after coapplication onto bare skin. Jun 2002

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